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THE EMBRYOLOGY OF THE ATLANTIC SALMON (SALMO SALAR LINNAEUS)[1]

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Abstract

Series of eggs collected at various Canadian Atlantic Coast Hatcheries from 1934 to 1940 were used as the bases for a survey of the embryology of the Atlantic salmon (*Salmo salar* L.) from fertilization to hatching. Early developmental stages from cleavage to the delineation of the embryonic shield are described. Somite formation commences when the embryonic axis is between 1 and 2 mm. in length, and is complete shortly after closure of the blastopore when 60 somites are evident at an embryonic length of 6 mm. Following this the embryo takes on a progressively more fish-like form until hatching.

The temperature of the water during most of the incubation period is relatively constant (0.5° C. to 1° C.) and the embryonic length data when plotted over this period fit the requirements for straight-line curves. The variation in the thermal units required to reach the same stage in different series indicates that their validity as criteria for determining comparable stages in embryonic development is doubtful.

Periods of greatest mortality in development occur during cleavage and blastoderm formation to the closure of the blastopore and at hatching.

Introduction

The importance of the Atlantic salmon (*Salmo salar* Linnaeus) as a food and game fish has long been unquestioned and its artificial propagation in federal hatcheries has been conducted on a major scale for many years. Nevertheless most scientific investigations upon this species have been concerned with its natural history and behaviour and with the growth, development, and care of the young fish. Shaw (39, 40) described briefly the development of the salmon from fertilization of the ovum to the age of two years. His (15, 16, 17), Ziegler (47), and Ryder (36) dealt in some detail with the early embryology of the salmon. Riddle (33) summarized the development of the Chinook salmon (*Oncorynchus tschawytscha*). A number of investigators, Lereboullet (25), Oellacher (27, 28, 29), Götte (7), Klein (19), Henneguy (13), Jablonovskij (18), and Kopsch (20, 21, 22), have dealt with early stages in the embryology of various species of trout. Price (30, 31, 32) made a comprehensive survey of development to hatching in the lake whitefish, *Coregonus clupeaformis* (Mitchill), a closely allied form with an egg similar to that of the salmon and requiring a prolonged incubation period.

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The present paper presents a general survey of the embryology of *Salmo salar* L., from fertilization to hatching, considered in relationship to both time and temperature factors as existing in a typical hatchery. Comparison is made with actual mortality percentages experienced at various developmental intervals. Such a survey has been undertaken not only to serve as a basis for future study, but also in the hope that it may yield information useful to those interested in various hatchery problems relative to salmon propagation.

Materials

The eggs of *Salmo salar* L. have an extended period of incubation, usually lasting from early or mid-November to the first or second week in May. Such a prolonged period permits collections of large series of eggs in closely graduated stages. During the winter of 1934-1935 one series (I) was collected at the St. John Fish Hatchery, St. John, N.B., daily from fertilization on Nov. 7 to Nov. 22, and thence weekly to July 26, 1935. Hatching took place between May 3 and May 10, with an approximate incubation period of 181 days. Temperature readings taken twice daily during this period varied from a minimum of 0.56° C. (33° F.) to a maximum of 7.2° C. (45° F.), but were constant at 0.56° C. from Dec. 7 to Apr. 20. A second series (II), also collected in 1934-1935, consisted of eggs reared at the Atlantic Biological Station Hatchery, St. Andrews, N.B. Samples were taken every two hours during the first 48 hr. following fertilization on Nov. 2, then daily to Nov. 16 and weekly thereafter until the supply was exhausted on Feb. 1. Development was considerably more rapid than in the former series since the temperature varied from a minimum of 2.2° C. (36° F.) to a maximum of 10° C. (50° F.). A third and more comprehensive series (III) was collected in the winter of 1939-1940 at the Birch Cove Hatchery, St. Andrews, N.B. It consisted of daily samples from fertilization on Nov. 17 to embryos apparently ready to hatch on May 20 (185 days). The temperature varied from a minimum of 0.5° C. (32.9° F.) to a maximum of 7.5° C. (45.5° F.), and was practically constant from 0.56° to 1.1° C. (33° to 34° F.) between Nov. 20 and Apr. 16.

The eggs were fixed in Bouin's fluid and preserved in 70% alcohol; or in Davidson's fluid, a formol–glycerine–alcohol–acetic-acid mixture. The fixation in the latter prevented the yolk from becoming brittle and was consequently more satisfactory for material that required sectioning than that in Bouin's.

Designation of Stages

Since development is a function of both temperature and time, Wallich (42) designated the age of fish embryos by what he termed "thermal units." One thermal unit (*t. u.*) corresponds to a temperature of 1° F. above 32° for a period of 24 hr., or is really a Fahrenheit-calorie–day. For example a mean temperature of 37° F. for one day yields five thermal units. It is thus possible, when the incubation period for any series is known in days, as

well as the mean daily temperature, to calculate the thermal units, thus expressing the age of an embryo as the sum of both time and temperature. This relationship has been found to hold within certain limits especially in Series I and III where the temperature ranges were similar, and will be used together with the age in days in the subsequent descriptions. Once the embryonic axis has been delineated the length of embryo and the number of somites will also be designated.

Characteristics of the Egg

The unfertilized egg of *Salmo salar* is spherical and varies from 5.5 to 6.0 mm. in diameter. It is covered by a heavy, translucent, somewhat elastic capsule, the chorion or zona radiata, which is produced in the ovary by follicular cells surrounding the egg during its formation. The chorion (Fig. 4, A)* is pierced by numerous very fine canals (1μ diameter) that run through it perpendicularly from its outer to its inner surface. These canals are so numerous that a surface view of the membrane (Fig. 4, B) appears marked by closely set fine dots representing the ends of the canals similar to those described by Henneguy (13) in the trout and by Becher (3) in various salmonoids. Ziegler (47) describes a funnel-shaped micropyle situated in an indentation on the surface of the chorion.

The bulk of the egg is composed of a spherical globule of somewhat amber-coloured yolk. It is a semifluid, viscid mass in the living egg (47) and free to revolve in the perivitelline space. A cap of pinkish-orange oil globules is accumulated at the upper surface of the yolk and occupies one-third to one-quarter of the diameter of the egg. It is 1 to 2 mm. deep at the centre. Similar though smaller globules are also irregularly scattered in a single layer at the margin of the yolk. A very thin cortical lamina of granular protoplasm is distributed over the entire surface of the yolk but is more concentrated in one region, the germinal disk or blastodisk.

Fertilized Egg

Following fertilization the blastodisk, although somewhat irregular in outline, becomes more evident and rests at the upper pole of the yolk upon the oil cap (Fig. 5). The latter has become more concentrated and fewer globules are scattered around the peripheral border of the yolk. Fig. 35 represents a transverse section through the lenticular blastodisk illustrated in Fig. 5, indicating the protoplasm fading out at the periphery into the thin cortical layer covering the yolk mass. Small, darkly-staining particles occur in the lower cytoplasmic layer and are probably yolk granules as described by His (16). Prior to the first cleavage, the blastodisk becomes elevated centrally and more or less definitely demarcated peripherally from the yolk by a ridge. It now measures 1.2 to 1.3 mm. in diameter.

* *Figs. 4 to 39 show the development of Salmo salar L.*

Cleavage

Two-celled Stage—(Series II, A. B. S.[1] Incubation period 13 to 15 hr.)

The first cleavage furrow is a vertical one and generally commences at the centre of the disk and extends transversely across it to either side (Fig. 6). Occasionally it begins at one side and progresses toward the opposite margin. The resultant cells or blastomeres are usually of equal size but sometimes one is considerably smaller than the other. This segmentation produces an oval disk with the longer axis perpendicular to the first cleavage plane.

Four-celled Stage—(Series II, A. B. S. Incubation period 19 hr.)

The second cleavage furrow is also meridional and commences at the centre of the disk at right angles to the first furrow. It is first observable 17 hr. after fertilization and is completed two to four hours later. The resultant four cells or blastomeres (Fig. 7) are usually equal in size, but if the first furrow has produced unequal cells, two are small, two large. Both cleavages extend to the lower level of the blastodisk but do not penetrate the yolk.

Eight-celled Stage—(Series II, A. B. S. Incubation period 23 hr.)

The blastodisk in this stage is still somewhat longer than broad and shows eight blastomeres arranged as four pairs (Fig. 8). The third division consisted of two transverse furrows, parallel to the first cleavage furrows, one through each pair of the first four cells. All eight-celled blastodisks are not elongated, although the majority show this pattern. Occasionally there are two tiers of cells, four smaller ones appearing to rest on four larger basal ones; or the eight cells are disposed symmetrically about a central point. Apparently in these instances the third furrow was a circular one at right angles to the first and second. It was first observable at 21 hr., completed in half the eggs by 23 hr., and in all by 25 hr., at a temperature of 8° C.

Sixteen-celled Stage—(Series II, A. B. S. Incubation period 33 hr., 19 *t.u.*)

The fourth cleavage furrow seems to be parallel to the second resulting in a one-layered disk (Fig. 9). The latter is losing its bilaterality and becoming rounded, although lobulated owing to the curvature of the individual marginal cells.

Thirty-two-celled Stage—(Series II, A. B. S. Incubation period 37 hr., 21 *t.u.*)

Fig. 10 illustrates a blastodisk approaching the 32-celled condition, and indicates the typical rounding up of the cytoplasmic mass. Occasionally the blastodisk at this stage is distinctly dumb-bell–shaped in surface view. Riddle (33) attributes this condition to a first furrow that is excessively deep in comparison to the succeeding ones. Transverse sections indicate that although only 32 cells may be evident in surface view, irregular horizontal cleavages have frequently resulted in a two-layered blastodisk.

[1] *A. B. S. = Atlantic Biological Station Hatchery.*

Later Segmentation

Through further cleavages the cells of the blastodisk become reduced in size, and arranged in first two, then subsequently more layers. A transverse section through a disk showing approximately 40 cells in surface view is illustrated in Fig. 36 (Series II, A. B. S. Nov. 4, 1934; incubation period 41 hr., 23 *t.u.*). It is three cells deep through the centre but shallower at its margin. The superficial cells are in active mitosis, are considerably broader than deep, and rather closely adherent to one another laterally, indicating the initiation of the formation of the epidermic stratum. The internal cells are loosely arranged and separated by irregular intercellular spaces representing the primordium of the segmentation cavity, as described by His (16) and by Kopsch (22) for the trout and Riddle (33) for the Chinook salmon. Ziegler (47) attributes the formation of the cavity to the fusion of the inner vacuolated ends of the blastomeres. It is distinctly different from the single continuous ventral one typical of many teleostean embryos, such as *Serranus* (43).

The marginal cells of the disk are elevated from the surrounding germinal material. The periblast is appearing as a distinct layer of granular protoplasm upon the surface of the yolk and is permeated by a few free nuclei. Darkly staining yolk granules are numerous in the central cells overlying the central region of the periblast, as well as in the vicinity of the periblast itself.

As cleavage continues the blastodisk becomes progressively divided into smaller cells or blastomeres and the whole structure is somewhat more prominently raised above the yolk surface (Fig. 11, Series II, A. B. S., Nov. 4, 1934; incubation period 43 hr., 25 *t.u.* and Fig. 12, Series II, A. B. S., Nov. 4, 1934; incubation period, 47 hr., 26 *t.u.*).

The outlines of individual blastomeres gradually become indistinct on the surface owing to the rapid cell division. This is indicated in Fig. 13, which represents an egg incubated for five days at an average temperature of 8.4° C., 77 *t.u.* (Series II, A. B. S., Nov. 7, 1934). The whole disk is commencing to spread out over the yolk surface. In transverse section the blastomeres are observed to occur in irregular strata of 10 to 12 cells, with small intermediate spaces representing the segmentation cavity or blastocoele. Ziegler (47) illustrates a rather large eccentrically-placed segmentation cavity of diameter 0.5 mm. and height 0.02 mm. in a blastoderm of 1.5 mm. diameter. This observation has not been corroborated in the present study and may have appeared through a contortion in sectioning.

The first slight cellular differentiation was previously noted in surface cells. In the multicellular disk, the external cells are continuous with one another and somewhat columnar in sectional area (Fig. 37, Series II, A. B. S., Nov. 6, 1934; incubation period four days, 59 *t.u.*; average temperature 8.4° C.). This so-called epidermic stratum forms a thin covering membrane and differs from the underlying loosely arranged spherical cells. It fuses with the granular periblast lying superficially on the yolk. The periblast itself extends completely under the blastodisk and immediately beyond its margin. It consists of rather large nuclei scattered throughout a cytoplasmic syncytium.

As cell division progresses, the blastomeres become correspondingly smaller in diameter and the blastoderm commences to spread over the surface of the yolk. Fig. 14 represents such a blastoderm, 1.7 mm. in diameter (Series II, A. B. S., Nov. 8, 1934; incubation period six days, 88 *t.u.*; average temperature 8.6° C.). The extent of the marginal periblast is evident as a granulation over the yolk surface at the edge of the blastoderm.

Formation of the Germ Ring and Subgerminal Cavity

As rapid segmentation proceeds, the blastoderm changes in shape and arches over the curvature of the yolk. A heavy band of cells is formed around the margin and encloses a lighter central area. This peripheral band constitutes the germ ring. It is slightly thickened at the point where the development of the embryo commences i.e. the embryonic bud or primordial shield (Fig. 15; Series I, St. John Hatchery, Nov. 19, 1934; incubation period 12 days, 84 *t.u.*; average temperature 4° C. and Fig. 16; Series I, St. John Hatchery, Nov. 22, 1934; incubation period 15 days, 99 *t.u.*; average temperature 3.9° C.).

As the blastoderm increases in diameter, it no longer appears lenticular in section (Fig. 36; Series II, A. B. S., Nov. 4, 1934; 41 hr. after fertilization, 23 *t.u.*; temperature 7.5° C.) and flattened on its lower surface (Fig. 37; Series II, A. B. S., Nov. 6, 1934; incubation period four days, 59 *t.u.*; average temperature 8.4° C.) but becomes concave to enclose the yolk sphere. This change, His (16, 17) has shown, is brought about not only by an active peripheral migration of the loose internal cells below the epidermic stratum, but also by a fusion of the intercellular spaces to form a subgerminal cavity. This elevates the blastoderm from the yolk except in the marginal zone, where the former is supported by individual groups of cells. Fig. 38 (Series II, A. B. S., Nov. 10, 1934; incubation period eight days, 127 *t.u.*; average temperature 8.4° C.), illustrates the early segmentation or subgerminal cavity lying slightly eccentrically between the central periblast and the overlying blastomeres and just within the germ ring.

On one side of the germ ring in the region of the embryonic bud or primordial shield (Figs. 15, 16) there occurs a heavy mass of compact undifferentiated cells. Its posterior margin constitutes the dorsal lip of the blastopore. An elongated cleft in this cellular mass extends anteriorly toward the subgerminal cavity, separating off a lower layer of cells, the primitive entoderm, which appears as a wedge-shaped lamina, one cell in depth medially, three to five cells peripherally. Götte (7) described the formation of this primitive entoderm for the trout, and His (16) for the salmon, as being brought about by centripetal ingrowth or invagination from the margin of the blastode m. The epidermic stratum does not take part in the ingrowth process, the latter being confined to the lower multicellular layer of the blastoderm. Oellacher (28), however, attributed its formation in the trout to a splitting of the original embryonic area into two layers by means of an elongated cleft. Ziegler (47) suggests the possibility that this may also occur in the salmon.

The germ ring continues to extend over the yolk surface and the embryonic shield commences to show some differentiation with a broadly elevated region anteriorly tapering to a narrower band at the dorsal lip of the blastopore (Fig. 17, Series II, A. B. S., Nov. 12, 1934; incubation period 10 days, 138 *t.u.*; average temperature 8° C.).

A sagittal section through the blastoderm at this stage of development is shown in Fig. 39 (Series II, A. B. S., Nov. 12, 1934; incubation period 10 days, 138 *t.u.*; average temperature 8° C.). Its extension over the yolk has been accompanied by a corresponding thinning of the roof over the subgerminal cavity. Cells of the epidermic stratum are becoming flattened. The entoderm extending forward now as a two-layered cellular tongue passes further into the subgerminal cavity, and appears as irregularly scattered cells beneath the epidermic stratum. The lower multicellular layer over the subgerminal cavity has become reduced to a few scattered cells. At the posterior end of the embryonic shield there is an undifferentiated caudal mass of cells bordering the dorsal lip of the blastopore.

In transverse sections at the stage illustrated in Fig. 17 and Fig. 39 the entoderm in the central axial portion of the embryo has fused into a line of cells, the anlage of the notochord.

Further overgrowth of the yolk by the blastoderm is accompanied by tissue differentiation in the embryonic area. A transverse section through the mid-region of the embryonic shield indicates that the ectoderm has become greatly thickened over the broad embryonic shield, as compared to the extraembryonic portion. The neural keel later develops from this thickening by compression of the cells from either side toward the median line (7, 17). Beneath this the chorda or primordium of the notochord has been differentiated as a solid rod of cells. Under the chorda, a thin layer of cells represents the entoderm, which will form the lining of the digestive tract and its various derivatives. Lateral to the notochord, the upper layer of the primitive entoderm has separated off as the mesoderm.

Investment of Yolk by Blastoderm and Early Differentiation of Embryo

Riddle (33) observed a curious relationship of the advancing germ ring over the yolk, to a circle of large oil drops on the surface of the latter, in the Chinook salmon. There seemed to be an indication that the ring of oil drops remained stationary while the germ ring passed over it. In a 2.4 mm. blastoderm (Fig. 15) a ring of large oil globules occurs beyond the margin of the germ ring, while at 2.85 mm. (Fig. 16), the innermost globules lie beneath it. At 3.75 mm. (Fig. 17) and 4.4 mm. (Fig. 18; Series II, A. B. S., Nov. 14, 1934; incubation period 12 days, 158 *t.u.*; average temperature 7° C.) the germ ring has extended to the edge of the circle and some of the globules occur at its inner margin. A study of later stages reveals the embryonic axis growing into the circle, the extraembryonic blastoderm advancing anteriorly beyond it. His (16, 17) showed in *Salmo salar*,

Wilson (43) in *Serranus*, and Price (30) in *Coregonus clupeaformis* that, during overgrowth or epiboly of the yolk by the blastoderm, that portion of the germ ring that constitutes the dorsal lip of the blastopore is relatively fixed, showing only slight backward growth. The anterior or ventral lip and lateral lips are most active in epiboly.

At the stage depicted in Fig. 18, the blastoderm has overgrown almost one-quarter of the yolk, and the anterior end of the embryonic shield has taken on a somewhat triangular appearance. The notochord is indicated externally by a somewhat clearer area along the embryonic axis extending forward about two-thirds the distance from the undifferentiated caudal mass at the dorsal lip of the blastopore to the anterior tip of the shield. The neural keel has become definitely established as a solid mass of cells in the mid-line and is composed of the nervous layer of ectoderm. At the anterior end the optic anlage have appeared as solid cellular masses projecting from either side of the keel. The mesoderm anteriorly is a flattened cellular layer while posteriorly the medial portions are commencing to show a cellular rearrangement to form a solid rosette charactistic of the future somite. The entoderm is undifferentiated and two or three layers thick medially but composed of only one layer laterally over the periblast.

When epiboly has proceeded until the blastoderm has covered approximately one-third of the yolk sphere, the embryonic axis has attained a length of 1.2 to 1.7 mm. and the mesoderm in the middle region has divided off one to two somites. By the time the yolk is nearly half enclosed by the overgrowing margin of the germ ring, the embryo has a length of 2.2 mm. and four mesodermal somites are visible externally (Fig. 19; Series I, St. John Hatchery, Nov. 29, 1934; incubation period 22 days; 129 *t.u.*; average temperature 3.4° C.). When the extraembryonic blastoderm has completely encircled the yolk it will constitute the yolk sac. The embryonic shield has become further elevated along its mid-line from the general surface of the blastoderm and the optic anlage are more definitely delineated in surface view. The neural keel is enlarged anteriorly as the primordium of the brain. There is a slight posterior extension of the embryo beyond the germ ring. This is probably due to some concrescence of the lateral lips of the blastopore and was previously noted by His (17) and Ziegler (47) in *Salmo salar*. This line of concrescence (43, 30) is homologous to the primitive streak of other forms and the mass of cells composing it may be termed the caudal prominence or mass (Figs. 28 and 29; Series I, St. John Hatchery, Nov. 29, 1934; incubation period 22 days; 129 *t.u.*; average temperature 3.4° C.). A slightly oblique sagittal section of a four-somite embryo (Fig. 29), just to one side of the mid-line, shows Kupffer's vesicle lying between the caudal prominence and the yolk. Its walls are one cell in thickness and the notochord extends forward from its anterior extremity. The nervous ectoderm constituting the neural keel or cord is thickened, especially anteriorly, although the epidermic stratum still remains flattened and composed of a single cellular layer. In transverse sections, the median portion of the keel is observed to be growing deeper, the

lateral portions thinner, probably, as suggested by Wilson (43), owing to cell migration.

The embryo shows considerable advances in differentiation when the blastoderm has covered two-thirds of the yolk sphere (Figs. 20 and 30; Series III, Birch Cove Hatchery, Jan. 24, 1940; incubation period 68 days; 109 *t.u.*; average temperature 1.5° C.). Seventeen pairs of mesodermal somites are present. The optic primordia have become optic vesicles by the appearance of slightly oblique internal clefts that connect directly with the anterior or cerebral ventricle. The latter has been formed by the development of a slit in the previously solid neural keel. The brain region formed from the anterior end of the latter has expanded considerably over the posterior portion, which is destined to form the spinal cord. The brain is, however, not yet differentiated into its primary divisions. Olfactory placodes occur as thickened areas of nervous ectoderm on either side of the mid-line at the extreme anterior tip of the embryo. Auditory placodes have similarly appeared as solid thickenings of the nervous ectoderm immediately anterior to the first mesodermal somites.

Thirty somites (Fig. 31, 4.2 mm. embryo; Series III, Birch Cove Hatchery, N.B., Feb. 5, 1940; incubation period 80 days; 120 *t.u.*; average temperature 1.3° C.) are visible when the blastoderm has overgrown three-quarters of the yolk sphere, presenting the appearance of a large yolk plug. The brain has commenced to show differentiation into its three primary vesicles, the forebrain, midbrain, and hindbrain. The neural keel in this region has continued to grow ventrally and is partially embedded in the yolk. Ventricles are present in the brain, the fourth extending somewhat laterally midway between the optic and auditory anlage. The optic anlage had previously become hollow structures or optic vesicles lying lateral to the forebrain and connected anteriorly with the anterior end of the latter. This connecting portion or optic stalk is also hollow serving as a free passageway from the brain ventricle into the optic vesicle. The lateral or retinal wall of the optic vesicle has become thickened, while the medial or future pigmented wall has become thinner, measuring only about one-quarter the width of the former. The ectoderm overlying the lateral wall of the optic vesicle has become thickened forming a placode, the anlage of the lens. It has already begun to press upon the lateral wall of the vesicle causing it to become concave, and thus transforming it into the optic cup. The auditory placodes have sunken deeply below the surface and have almost become cut off as hollow sacs, the auditory vesicles. The olfactory placodes have undergone no marked change, other than a deepening of the pit-like depressions.

The yolk plug is approximately 1 mm. in diameter, when 35 somites are visible along the embryonic axis (Fig. 21, 4.4 mm. embryo, Series III, Birch Cove Hatchery, Feb. 8, 1940; incubation period 83 days; 124 *t.u.*; average temperature 1.3° C.). Other than an increase in the length of the body and more extensive epiboly, this embryo resembles the 30-somite one previously described.

When 40 somites (Fig. 32; 4.8 mm. embryo; Series I, St. John Hatchery, Dec. 13, 1940; incubation period 36 days; 164 *t.u.*; average temperature 2.5° C.) have been differentiated the blastopore has closed completely. Occasionally its former position is indicated by a slight furrow on the yolk sac just posterior to the caudal region. Differentiation has been rapid about the period of closure of the blastopore. The embryo is now prominently elevated above the yolk surface, without torsion. It extends around one-third the circumference of the yolk sphere. The olfactory placode has become a deepened pit. The lens has separated from the surface ectoderm and has migrated into the mouth of the optic cup. The choroid fissure is evident as a triangular slit in the ventral margin of the cup. The optic stalks are in direct communication with the third ventricle. The auditory vesicle has enlarged considerably and is completely separated from the surface ectoderm. Its wall consists of a thickened layer of columnar epithelial cells concentrically arranged, but deeper on the medial and ventral wall than on the lateral aspect. The brain has increased in size and its walls have become thicker. Three outgrowths have been formed from the forebrain; one is directed dorsally and forward, the cerebrum; another is directed upward and backward from the roof of the forebrain forming the pineal body. It extends to the surface ectoderm, at the posterior margin of the cerebrum. The third outgrowth, the infundibulum, has appeared as a backwardly directed ventral projection from the forebrain, toward the end of the notochord. The midbrain is now divided into two distinct optic lobes, and the hindbrain is differentiated into the metencephalon (cerebellum) and the myelencephalon (medulla oblongata). The walls of the brain stem have thickened and the fourth ventricle is enlarged.

A dorsal fin fold extends from the head region posteriorly, surrounding the caudal region as the forerunner of the caudal fin. Pectoral fins have appeared as lateral extensions of the body wall at the level of the first somites. The notochord is prominent. Its anterior end is flexed slightly ventrad and tapers to a point at the anterior margin of the medulla oblongata. Its posterior end is broader and merges into the undifferentiated tissues of the tail. The first indication of vacuolation is evidenced by the peripheral migration of nuclei throughout its course. The mesodermal somites in lateral view are elongated dorsoventrally and each is bent at its middle, the apex of the angle formed by the bend, pointing anteriorly. A transverse section of the embryo at the level of an anterior mesodermal somite shows a few cells along the ventromesial border taking on the mesenchymal characteristics of the sclerotome. The intermediate cell mass has migrated ventrally and forms irregular masses. The lateral plate mesoderm is divisible into somatic and splanchnic layers, which are separated by the coelom.

The entoderm is a simple unfolded layer overlying the yolk throughout the body, anteriorly to the branchial region. The pharynx has been formed by an elevation of the entoderm medially and a folding under laterally, resulting in a flattened tube. Three pairs of heavy dorsolateral branchial folds form

on either side of the pharynx somewhat anterior and ventral to the ear. They project distally as pouches to meet plates of surface ectoderm on the side of the head, marking the future location of the first three clefts. The pharynx terminates just anterior to the tip of the notochord. The heart is indicated by the appearance of folds of mesoderm ventrolateral to the branchial folds that migrate down from either side of the pharynx and join to form a tube enclosed in the pericardial cavity. Cells on the medial border of the folds elongate and between these lie irregular cells that will constitute the endothelial lining of the heart.

Later Development of the Embryo

Following completion of the yolk sac, resulting from epiboly of the extra-embryonic blastoderm, the body enlarges rapidly. The tail is distinctly raised off the surface of the yolk and has become flattened from side to side. (Fig. 22; 5.8 mm. embryo, 58 somites; Series I, St. John Hatchery, Dec. 20, 1934; incubation period 43 days, 170 *t.u.;* average temperature 2.5° C.; Fig. 33; 6.0 mm. embryo, 60 somites; Series III, Birch Cove Hatchery, Feb. 20, 1940; incubation period 95 days, 136 *t.u.*; average temperature 1° C.). The embryo, with continued growth, has acquired a more fish-like appearance. It lies prominently over the yolk sac. The brain has increased greatly in size, more especially by growth of the optic lobes and cerebellum. The optic lobes have extended dorsally, posteriorly, and laterally until they overhang the infundibulum. The infundibulum still remains ventral to the midbrain, but its posterior limit is the anterior end of the cerebellum, which is now a prominent transverse plate. The medulla oblongata has expanded laterally by expansion of the fourth ventricle, which appears triangular in shape from dorsal view. Posterior to the auditory vesicle the medulla rounds up into the tubular nerve cord, which extends to the tail region. It disappears into the caudal mass. The nasal pits have become deeper. The cavity of the original optic vesicle is obliterated and its two walls have been brought in contact. The optic cup now appears as a flattened sphere with a large cavity almost filled by the lens. The wall of the cup is interrupted ventrolaterally by the triangular choroid fissure. The pectoral fins are well developed. The median fin-fold is continuous around the tail from the dorsal to the ventral median line. Vacuolation of the notochordal cells has become more distinct.

A study of transverse sections indicates that the fore-gut terminates somewhat posterior to the orbit. The remainder of the gut is a straight, closed tube ending in a loose mass of cells, marking the location of the future anus, approximately one-third of the body length anterior to the tip of the tail.

Differentiation of Mesodermal Somites

The formation of the first paired mesodermal somites has been observed when the embryonic axis is 1.2 mm. in length (Fig. 1). At an embryonic length of 5.8 to 6 mm., the adult number of 60 somites is present. Examination of the graph prepared from somite counts on Birch Cove Hatchery

material (Series III) indicates that somite formation is at first slow (from 1 to 2 mm. embryonic length) then progessively becomes more rapid until the adult number is attained.

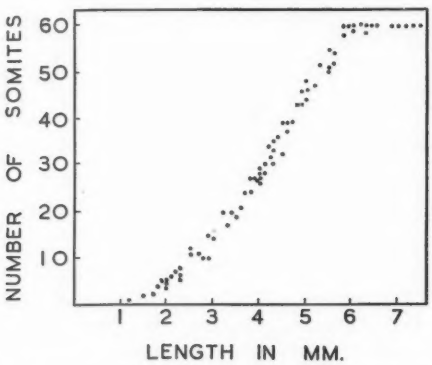


FIG. 1. *The relation between the length in millimetres of the embryo of Salmo salar L. and the number of mesodermal somites present.*

Length in millimetres	*Range of somite numbers*
1 – 2	*1 – (4 – 5)*
2 – 3	*5 – (9 – 11)*
3 – 4	*10 – (26 – 29)*
4 – 5	*28 – (44 – 48)*
5 – 6	*46 – (59 – 60)*

Price (30) showed for the whitefish that once formation of the paired somites is initiated an increase in their number apparently occurs at a rate directly proportional to the number of thermal units to which the embryo has been subjected. The complete number of somites is differentiated before the embryo has completed the first half of its incubation period.

For the salmon in Series I, St. John Hatchery, N.B., the complete number of somites was developed by Dec. 20, 1934, i.e. with an incubation period of 43 days at an average temperature of 2.5° C. This is approximately the end of the first quarter of the total incubation period (181 ± 3) days and equivalent to 170 thermal units or 38% of those required to bring about hatching (436 *t.u.*). For Series III, Birch Cove Hatchery, N.B., the complete number of 60 somites was first present on Feb. 20, 1940, with an incubation period of 95 days, at an average temperature of 1° C. This is approximately one-half of the total incubation period (185 +) days and equivalent to 136 thermal units or 29% of those that would be required to bring about hatching (477 + *t.u.*). The variation in the thermal units required to reach the same status in these two series throws some doubt on their validity as criteria for determining comparable stages in embryos. Discrepancies may, however, be accounted for at least in part by differences in amounts of latent heat contained in different waters at the freezing point of water and also to some extent by differences in thermometers.

Development Prior to Hatching

During the latter three-quarters of the incubation period in Series I, St. John Hatchery, N.B., or the latter half of the period in Series III, Birch Cove Hatchery, N.B., development of the salmon embryo as far as external characteristics are concerned involves largely an increase in body size accompanied by a decrease in yolk material, and the appearance of pigmentation first in the eye then lightly over the dorsal body surface (Figs. 23 to 27).

The embryo illustrated in Fig. 23 (8.0 mm. embryo; Series I, St. John Hatchery, Jan. 10, 1935; incubation period 64 days, 192 *t.u.*; average temperature 1.8° C.) is entirely lacking in pigmentation, and usually lies with slight torsion on the upper surface of the yolk sac. The latter is beginning to show indications of heavy vascularization. The optic lobes are prominent and somewhat ovoid. Fig. 24 (10.5 mm. embryo; Series I, St. John Hatchery, Feb. 1, 1935; incubation period 86 days, 214 *t.u.*; average temperature 1.7°C.) represents a somewhat longer embryo. The tissues have become thickened and opaque and virtually obstruct the appearance of the ears through the body surface. Three gill slits are evident. Pigment appears in the eyes when the embryo has grown to extend approximately three-quarters of the distance around the yolk sac (Fig. 25; 13.5 mm. embryo; Series I, St. John Hatchery, N.B., Mar. 15, 1935; incubation period 128 days, 256 *t.u.*; average temperature 1.2° C.).

As the embryo takes on the appearance of the young fish or alevin (Fig. 26; 15.8 mm. embryo; Series I, St. John Hatchery, N.B., Apr. 12, 1935; incubation period 156 days, 284 *t.u.*; average temperature 1.1° C.) the eyes become more deeply pigmented and fine stellate melanophores are present over the brain and dorsal trunk musculature. The pectoral fins are no longer fleshy protuberances but are thin folds in which supporting rays have developed.

Body pigmentation increases as the embryo forms almost a complete circle on the yolk (Fig. 27, 18.6 mm. embryo; Series I, St. John Hatchery, N.B., Apr. 26, 1935; incubation period 170 days, 310 *t.u.*; average temperature 1.1° C.). Rays arise in the dorsal, caudal, and pectoral fins. Before hatching the embryo completely encircles the yolk with the tail extending past the head sometimes as far as the posterior margin of the operculum, and the bases of the pectoral fins. Pelvic fins appear as fleshy buds at the posterior junction of the yolk sac and the ventral trunk musculature.

Just after hatching (Fig. 34; total length 22 mm.; Series I, St. John Hatchery, N.B., May 10, 1935; incubation period 181 $\pm$ 3 days, 470 *t.u.*; average temperature 1.6° C.) the larva or alevin bears a heavy yolk sac, somewhat triangular in shape from the lateral aspect. The eye is deeply pigmented. Scattered melanophores occur over the dorsal surface of the head and body musculature as far as the lateral line. Only a few appear ventral to it.

The dorsal fin-fold is becoming differentiated. An anterior elevation supported by 11 fin rays marks the location of the dorsal fin of the adult; a

posterior one, the location of the adipose fin. A similar ventral fold, posterior to the anus, and supported by nine rays will develop into the anal fin. The pelvic fins are still small lateral fleshy protuberances midway between the pectoral fins and the anus. The gill slits are entirely covered by the oper-

TABLE I

GROWTH OF BLASTODERM OF *Salmo salar* L., ST. JOHN HATCHERY, N.B., 1934-35

Date	Age		Diameter (mm.)
	Days	Thermal units	
Nov. 8	1	11.5	1.30 × 1.25
Nov. 9	2	24.0	1.50
Nov. 10	3	36.5	1.60
Nov. 11	4	43.0	1.80
Nov. 12	5	49.5	1.85
Nov. 13	6	54.5	1.85
Nov. 14	7	58.0	1.90
Nov. 15	8	61.5	1.90
Nov. 16	9	65.0	2.00
Nov. 17	10	68.5	2.00*
Nov. 18	11	73.0	2.20
Nov. 19	12	78.5	2.40**
Nov. 20	13	83.5	2.50
Nov. 21	14	88.5	2.60
Nov. 22	15	93.5	2.80

* *First definite appearance of germ ring.*
** *Location of future embryonic axis (embryonic shield) indicated.*

TABLE II

GROWTH OF BLASTODERM OF *Salmo salar* L., BIRCH COVE HATCHERY, N.B., 1939-40

Date	Age		Diameter (mm.)
	Days	Thermal units	
Nov. 17	—	—	1.20
Nov. 20	3	11.7	1.20
Nov. 23	6	17.4	1.22
Nov. 26	9	22.2	1.28
Nov. 29	12	25.9	1.29
Dec. 2	15	29.6	1.41
Dec. 5	18	40.8	1.47
Dec. 8	21	49.4	1.50
Dec. 11	24	53.0	1.54
Dec. 14	27	57.6	1.66
Dec. 17	30	61.7	1.95*
Dec. 20	33	66.7	2.35**
Dec. 23	36	71.7	2.72
Dec. 26	39	75.3	3.00
Dec. 29	42	78.8	3.05
Jan. 1	45	82.3	3.10

* *First definite appearance of germ ring.*
** *Location of future embryonic axis (embryonic shield) indicated.*

culum although a few gill filaments protrude at its dorsolateral margin. Sixty myomeres are present.

Rate of Growth

Following early cleavage, the blastoderm increases in diameter.

Measurements of the diameter of the blastoderms of the St. John Hatchery Series, 1934-35, for the first 15 days are given in Table I; those for the Birch Cove Hatchery Series 1939-40, for the first 45 days, in Table II. The former represent averages from 10 individuals, the latter from six. The increase in diameter is at first very gradual, until the germ ring has appeared[1]. It is much more rapid following this as active epiboly of the yolk by the blastoderm commences and the embryonic shield[2] develops (Figs. 2 and 3).

When the yolk has been one-quarter to one-half overgrown by the blastoderm, it is possible to measure the length of the embryonic axis with some accuracy. The average length measurements of 10 individuals from each weekly sample of the St. John Hatchery series from Nov. 29, 1934, to May 3, 1935, is given in Table III, and graphically represented in Fig. 2. For the Birch Cove Hatchery Series (Table IV), three specimens were measured daily from Jan. 12, 1939, to May 20, 1940, and individual points plotted in Fig. 3. Averages for the seventh day (Saturday) of each week are given in Table IV.

Examination of Figs. 2 and 3 shows that the embryonic length data, when plotted over the periods of constant temperature fit the requirements for

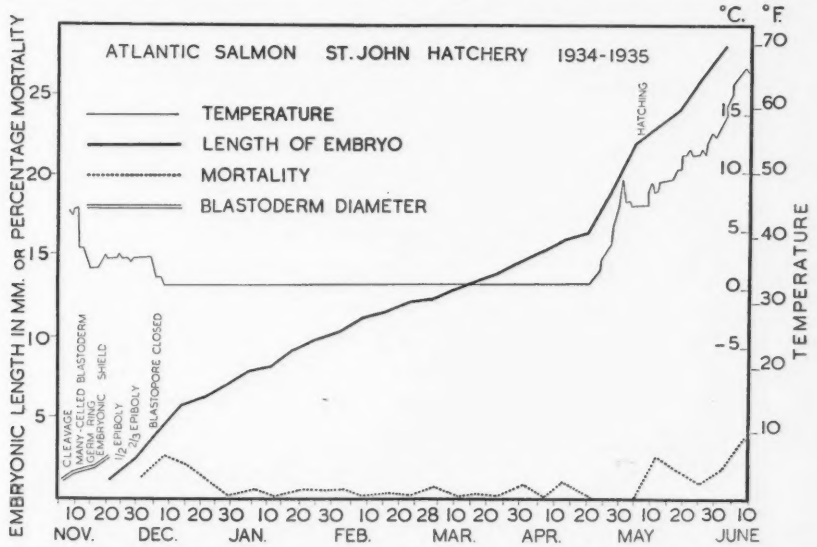


FIG. 2. *Graph indicating the growth in length of the embryo of Salmo salar L. and time (heavy line) at St. John Hatchery, N.B., 1934-35. Individual points were obtained by averaging measurements of 10 individuals.*

[1] *See first footnote, Tables I and II.*

[2] *See second footnote, Tables I and II.*

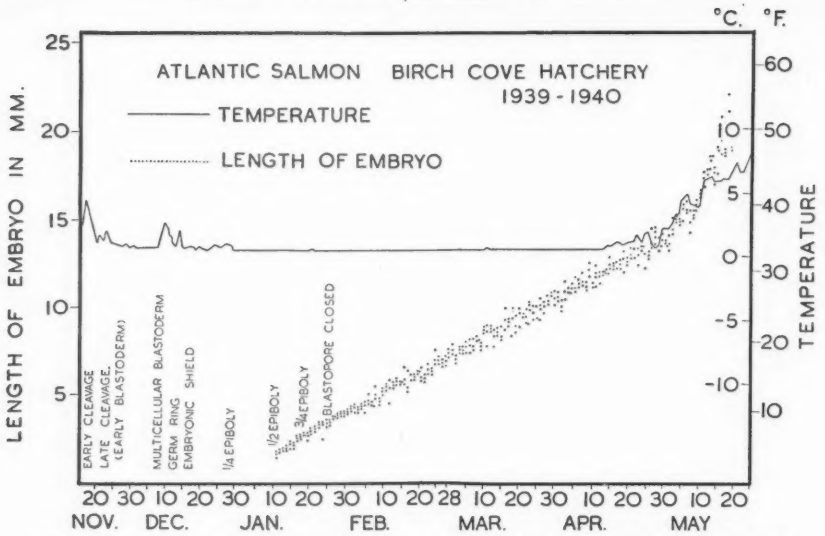


FIG. 3. *Graph indicating the growth in length of the embryo of Salmo salar L., at Birch Cove Hatchery, N.B., 1939-40. Growth curve is plotted by measurement of three individuals per day.*

TABLE III

GROWTH OF EMBRYONIC AXIS IN *Salmo salar* L.,
ST. JOHN HATCHERY, N.B., 1934-35

Date	Age		Length (mm.)
	Days	Thermal units	
Nov. 29	22	129	2.4
Dec. 6	29	156	4.2
Dec. 13	36	164	5.8
Dec. 20	43	171	6.2
Dec. 27	50	178	7.0
Jan. 3	57	185	7.9
Jan. 10	64	192	8.1
Jan. 17	71	199	9.2
Jan. 24	78	206	9.8
Feb. 1	85	214	10.3
Feb. 8	92	221	11.2
Feb. 15	99	228	11.5
Feb. 22	106	235	12.2
Mar. 1	113	242	12.4
Mar. 8	120	249	12.8
Mar. 15	127	256	13.4
Mar. 22	134	263	13.8
Mar. 29	141	270	14.6
Apr. 5	148	277	15.2
Apr. 12	155	284	15.9
Apr. 19	162	291	16.2
Apr. 26	169	310	18.8
May 3	176	384	22.0

NOTE:—*Incubation period: Nov. 7 to May 7 (to 10) = 181 ± 3 days.*
Average temperature: 1.4° C.
Minimum temperature: 0.56° C.
Duration of minimum temperature: Dec. 7 to Apr. 20 = 135 days.

TABLE IV

GROWTH OF EMBRYONIC AXIS IN *Salmo salar* L., BIRCH COVE HATCHERY, N.B., 1939-40

Date	Age		Length (mm.)
	Days	Thermal units	
Jan. 13	57	97	1.9
Jan. 20	64	105	2.8
Jan. 27	71	112	3.5
Feb. 3	78	119	4.2
Feb. 10	85	126	4.6
Feb. 17	92	133	6.0
Feb. 24	99	141	6.5
Mar. 2	106	149	7.5
Mar. 9	113	159	8.2
Mar. 16	120	167	8.5
Mar. 23	127	175	9.4
Mar. 30	134	183	10.9
Apr. 6	141	192	11.3
Apr. 13	148	203	11.7
Apr. 20	155	221	12.6
Apr. 27	162	243	12.9
May 4	169	294	14.5
May 11	176	367	16.1
May 18	183	452	19.5

NOTE:—*Incubation period: Nov. 17 to May 20+ = 185+ days.*
Average temperature: 1.4° C.
Minimum temperature: 0.5° to 1° C.
Duration of minimum temperature: Nov. 22 to Apr. 23 (except Apr. 16, 17, 19, 20) = 154 days.

straight-line curves using the Equation $y = ax + b$. Fitting by the method of least squares these equations were respectively: for the St. John Hatchery, $y = 0.08228x + 3.089$; for the Birch Cove Hatchery, $y = 0.10921x + 4.2819$. They indicate that while corresponding lengths are attained at very different times, yet the rate of development during this period of constant temperature is almost identical as shown by the close approximation of the slopes from the two equations.

At the St. John Hatchery (Fig. 2) with higher temperatures prior to Dec. 7, and after Apr. 20, the rate of growth is considerably more rapid than during the winter temperature range. Similarly at Birch Cove Hatchery (Fig. 3), after Apr. 23 the embryonic axis increases in length much more rapidly than in the previous constant low temperature period. Length measurements commencing on Jan. 12 maintain a straight line relationship to Apr. 27.

Scheminzky and Gauster (38) measured the length of the embryos of *Salmo lacustris* at approximately 10-day intervals to hatching (53 days) with a water temperature of 8° C. From the 1st to 10th, and from the 20th to the 80th days, the growth curves have approximately the same slope and are straight lines. From the 10th to 20th days development is more rapid and involves the period of overgrowth of the blastoderm over the yolk and the

early formation of the embryonic axis. This is similar to the period of rapid growth in *Salmo salar* Series I from Nov. 29 to Dec. 13 although the average temperature for that period (1.5° C.) was slightly above the average one (0.56° C.) for the subsequent four months.

Gray (8) found that the growth curve (wet weight) of embryos of *Salmo fario* incubated at 10° C. for approximately 55 days is markedly convex to the time axis. It then remains more or less linear until 80 days, after which it suddenly becomes concave until development is complete at 100 days after fertilization. Allen (1) has illustrated graphically a series of length measurements of *Salmo salar*, which appears to be a straight line for the 30 days prior to hatching (1.5 to 17.5 mm.) with an average temperature of 10.2° C. Since the Atlantic salmon material used in the present study was not subject to constant temperature at the beginning and at the termination of its incubation period its growth is scarcely comparable to that of the instances cited, other than that a straight-line relationship with time is exhibited during an interval following delineation of the embryonic axis and prior to hatching.

Mortality Periods

A number of investigators have observed that the susceptibility of fish eggs to external influences changes during the incubation period. Hein (11, 12) subjected eggs of the trout (*Salmo fario*) to various types of adverse conditions such as light, pressure, shock, heat, and cold, at different intervals after fertilization. He observed a gradual decrease in susceptibility to the closure of the blastopore (13 to 17 days) and a subsequent increase just prior or at hatching. Scheminzky *et al.* (37, 38) obtained similar results when eggs of the trout, *Salmo lacustris*, were exposed to electric currents. Higurashi and Nakai (14) using eggs of a Japanese fish, *Plecoglossus altivelis*, found them very susceptible to cold in early stages but they became more resistant after the appearance of the eye spots. Hata (9) using vibrations on the eggs of *Oncorhynchus masou*, obtained results that corroborated the observations of the previous workers. The lethal temperature of the developing eggs of the four-bearded rockling (*Enchelyopus cimbrius*) increases from fertilization to a maximum just prior to the closure of the blastopore, then decreases, before rising to a second high value immediately preceding hatching (2). Rollefsen (34, 35) determined the effect of mechanical agitation on the pelagic egg of the cod, and found that the rapid growth of the blastodisk over the yolk was accompanied by a corresponding decrease of the susceptibility. He attributed the increased resistance of the egg to the covering up of the yolk by the embryonal tissue. Allen (1) observed a susceptibility of the eggs of *Salmo salar* to mechanical shock during organogenesis and at the time of hatching. Worley (46) states for the mackerel that; "apparently the stages of germ ring formation and epiboly and the somite multiplication stages are critical periods in the development of the embryo." Bonnet (4) found for the cod, that at all temperatures an initial period of high mortality decreased with the closing of the blastopore and was followed by a period of low mortality until the

embryo was three-quarters the circumference of the egg membrances, when the death rate steadily increased up to hatching. Vernidub (41) showed that maximum mortality of *Salmo fontinalis* embryos, subjected to injurious agents such as high temperature, hydrogen, and potassium cyanide, occurred at the commencement of cleavage, during the formation of the axial organs and closing of the blastopore, as well as just prior to the appearance of eye pigmentation, when the anlage of the pectoral fins is first evident.

Since Series I was collected at the St. John Hatchery, N.B., weekly records of mortality percentages are available. These have been plotted graphically (Fig. 2) and are as follows:

Dec. 1	1.25	Feb. 23	—
Dec. 8	2.61	Mar. 2	0.78
Dec. 15	2.18	Mar. 9	—
Dec. 22	1.04	Mar. 16	0.13
Dec. 29	—	Mar. 23	—
Jan. 5	0.46	Mar. 30	0.09
Jan. 12	—	Apr. 6	—
Jan. 19	0.44	Apr. 13	0.09
Jan. 26	0.44	Apr. 20	—
Feb. 2	0.47	May 4	—
Feb. 9	—	May 11	2.49
Feb. 16	0.19		

The percentages are too low to warrant tentative conclusions with the possible exception of those at the commencement and at the termination of the incubation period. The first record of 1.25% on Dec. 1 includes all stages from fertilization through cleavage, germ ring formation, and partial overgrowth of the yolk. The closure of the blastopore with the accompanying development of the eye and ear, and the undercutting of the tail from the yolk surface by the tail fold, would seem to represent the termination of critical stages, with a loss of 2.61% during the week ending Dec. 8 and 2.18 for Dec. 15.

At hatching (May 7 $\pm$ 3) the mortality again rises to 2.49% for the week ending May 11. The hatching period appears to be a critical one perhaps owing to some defect in the enzyme (5, 6, 10, 24, 44, 45) that appears prior to hatching, or it may also be indicative of earlier faulty development that either prevents or interferes with the normal processes of hatching.

M'Gonigle (26), from studies of records for the Canadian Atlantic coast hatcheries, points out that the age of the parents has a bearing on the early mortality (prior to closure of the blastopore) and the later one (prior to hatching) in both salmon and trout. In young parents the major loss is the early one, and tends to decrease with the age of the parent, while the later mortality increases.

Summary

1. This paper is the result of a preliminary survey of the embryology of the Atlantic salmon (*Salmo salar* L.) from fertilization to hatching. It is based upon series of eggs collected at the St. John Hatchery, N.B., and the Atlantic Biological Station, St. Andrews, N.B., in 1934-35 and at the Birch Cove Hatchery, N.B., in 1939-40.

2. The salmon is typical of the teleosts in its general development. The eggs are approximately 5.5 mm. to 6 mm. in diameter, and have an extended incubation period usually from early November until early May. Oil globules in the yolk have a tendency to concentrate about the blastodisk.

3. Cleavage, the development of the early segmentation cavity as scattered interspaces between the blastodermic cells, the formation of the germ ring, the subgerminal cavity, the overgrowth of the blastoderm about the yolk, and the delineation of the embryonic shield are described.

4. Somite formation commences when the embryonic axis is between 1 and 2 mm. in length, and is complete shortly after closure of the blastopore when 60 somites are evident at an embryonic length of 6 mm.

5. Immediately prior to the closure of the blastopore the embryo lying in a straight line over the curvature of the yolk is 4.4 mm. long, or approximately one-fifth of its hatching length. Thirty-five somites are differentiated and the yolk plug is 1 mm. in diameter. The three primary brain lobes are distinct and the optic anlage is in the form of a cup into the cavity of which the lens placode projects. The auditory organ appears as a vesicle. The olfactory placode forms a shallow pit.

6. Following closure of the blastopore, the embryo gradually takes on a progressively more fish-like form until hatching. Externally this includes an increase in size; the appearance of pigmentation, first in the eye, then later over the general body surface more especially dorsal to the lateral line; and differentiation of the fins.

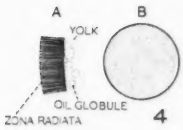
7. The variation in the thermal units required to reach the same status in the two comparable series, namely, St. John Hatchery, 1934-35, and Birch Cove Hatchery, 1939-40, throws some doubt on the validity of thermal units as criteria for determining precise stages in embryonic development.

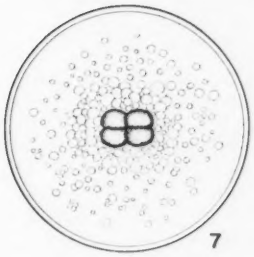
8. The rate of growth of the embryonic axis exhibited a straight line relationship with time during that part of the incubation period when the temperature was relatively constant (0.56° C., St. John Hatchery and 0.5° C. to 1° C., Birch Cove Hatchery).

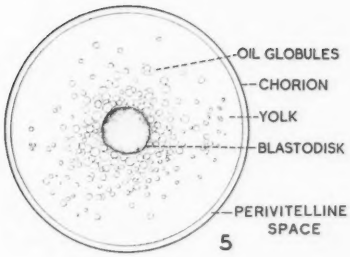
9. Periods of greatest mortality during development would seem to be during cleavage and blastoderm formation, up to the closure of the blastopore (i.e. during organogenesis), and at hatching.

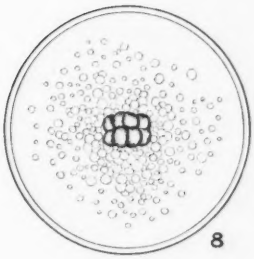
Acknowledgments

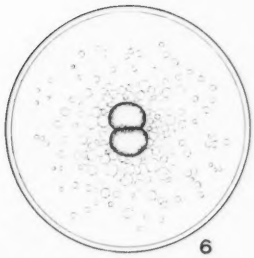
The author wishes to express her appreciation to the Fisheries Research Board of Canada for the use of this material, and especially to Dr. R. H. M'Gonigle who undertook the detailed task of its collection. To Dr. A. H. Leim, Director of the Atlantic Biological Station, St. Andrews, N.B., and to Professor A. D. Robertson, Head of the Department of Zoology, University of Western Ontario, special thanks are due not only for interest and suggestions concerning the problem, but also for so freely permitting use of all laboratory facilities.











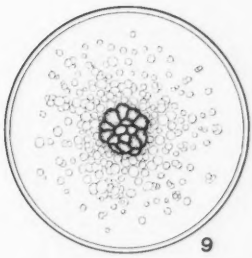


FIG. 4. *A. Transverse section through chorion (zona radiata) of advanced ovarian egg. Width 0.01 mm.*

B. Surface view of chorion showing ends of perpendicular canals.

FIG. 5. *Polar view of recently fertilized egg. Diameter of blastodisk 1.2 mm. Series II. A. B. S., Nov. 2, 1934. Seven hours after fertilization. Temperature 7° C.*

FIG. 6. *Surface view of egg in two-celled stage. Length of blastodisk 1.2 mm. Greatest width of each cell 1 mm. Series II. A. B. S., Nov. 3, 1934. Fifteen hours after fertilization. Temperature 8° C.*

FIG. 7. *Surface view of egg in four-celled stage. Length of blastodisk 1.25 mm. Width of blastodisk 1 mm. Series II. A. B. S., Nov. 3, 1934. Nineteen hours after fertilization. Temperature 8° C.*

FIG. 8. *Surface view of egg in eight-celled stage. Series II. A. B. S., Nov. 3, 1934. Twenty-three hours after fertilization. Temperature 8° C.*

FIG. 9. *Surface view of egg in 16-celled stage. Series II. A. B. S., Nov. 3, 1934. Thirty-three hours after fertilization. Temperature 8° C.*

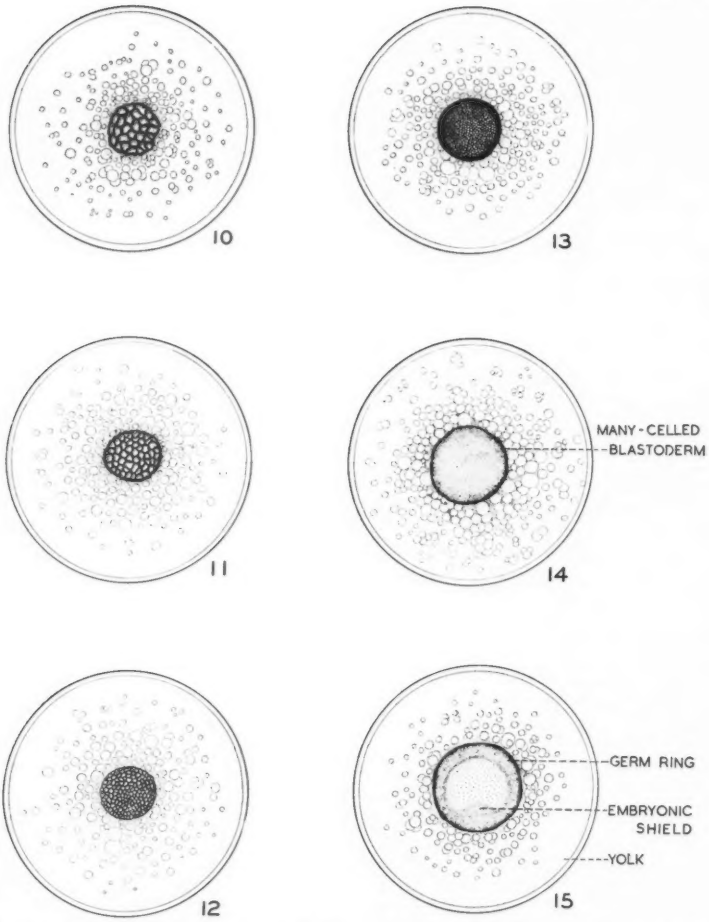


FIG. 10. *Surface view of egg in 32-celled stage. Diameter 1.4 mm. Series II. A. B. S., Nov. 4, 1934. Thirty-seven hours after fertilization. Temperature 7.5° C.*

FIG. 11. *Surface view of blastodisk of diameter 1.45 mm. Series II. A. B. S., Nov. 4, 1934. Forty-three hours after fertilization. Temperature 7.5° C.*

FIG. 12. *Surface view of blastodisk of diameter 1.45 mm. Series II. A. B. S., Nov. 4, 1934. Forty-seven hours after fertilization. Temperature 7.5° C.*

FIG. 13. *Surface view of blastodisk of diameter 1.55 mm. Series II. A. B. S., Nov. 7, 1934. Five days after fertilization. Average temperature 8.4° C.*

FIG. 14. *Surface view of blastoderm of diameter 1.70 mm. Series II. A. B. S., Nov. 8, 1934. Six days after fertilization. Average temperature 8.6° C.*

FIG. 15. *Surface view of blastoderm showing beginning of germ ring. Diameter through embryonic shield 2.4 mm. Series I. St. John Hatchery, Nov. 19, 1934. Incubation period 12 days, 84 t.u. Average temperature 4° C.*

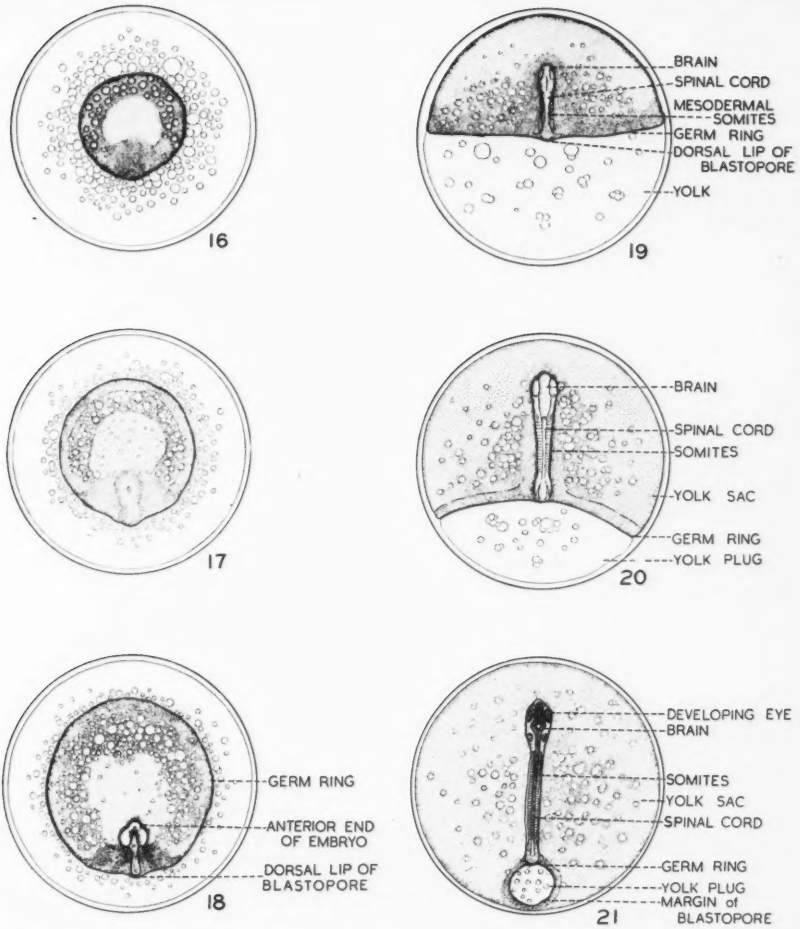


FIG. 16. *Surface view of blastoderm showing increasing diameter of germ ring. Diameter through embryonic shield 2.85 mm. Series I. St. John Hatchery, Nov. 22, 1934. Incubation period 15 days, 99 t.u. Average temperature 3.9° C.*

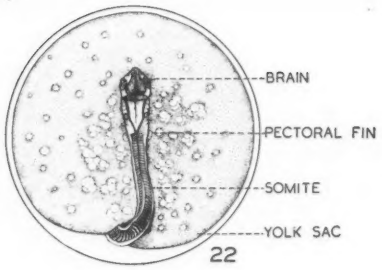
FIG. 17. *Surface view of blastoderm showing advanced germ ring. Diameter through embryonic shield 3.75 mm. Series II. A. B. S., Nov. 12, 1934. Incubation period 10 days, 138 t.u. Average temperature 8° C.*

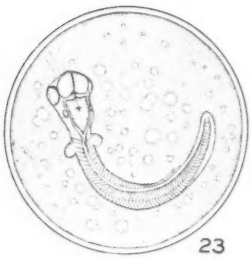
FIG. 18. *Surface view of blastoderm showing first external indication of differentiation of anterior end of embryonic shield. Greatest diameter of blastoderm 4.4 mm. Series II. A. B. S., Nov. 14, 1934. Incubation period 12 days, 158 t.u. Average temperature 7° C.*

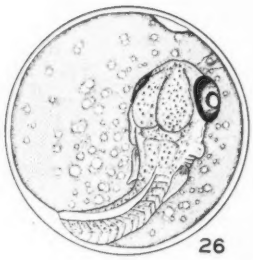
FIG. 19. *Surface view of developing egg. The blastoderm has overgrown half of the yolk sphere. Length of embryonic axis 2.12 mm., five somites. Series I. St. John Hatchery, N.B., Nov. 29, 1934. Incubation period 22 days, 129 t.u. Average temperature 3.4° C.*

FIG. 20. *Surface view of egg. The blastoderm has overgrown two-thirds of the yolk sphere. Length of embryonic axis 3.3 mm., 17 somites. Series III. Birch Cove Hatchery, N.B., Jan. 24, 1940. Incubation period 68 days, 109 t.u. Average temperature 1.5° C.*

FIG. 21. *Surface view of developing egg with narrow yolk plug. Total length of embryo 4.4 mm., 35 somites. Series III. Birch Cove Hatchery, N.B., Feb. 8, 1940. Incubation period 83 days, 124 t.u. Average temperature 1.3° C.*







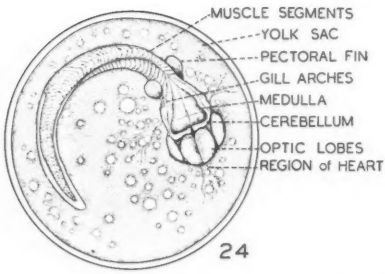


FIG. 22. *Surface view of developing egg. Length of embryo 5.8 mm., 58 somites. Series I. St. John Hatchery, N.B., Dec. 20, 1934. Incubation period 43 days, 170 t.u. Average temperature 2.5° C.*

FIG. 23. *Surface view of developing egg. Length of embryo 8.0 mm. Series I. St. John Hatchery, N.B., Jan. 10, 1935. Incubation period 64 days, 192 t.u. Average temperature 1.8° C.*

FIG. 24. *Surface view of developing egg. Length of embryo 10.5 mm. Series I. St. John Hatchery, N.B., Feb. 1, 1935. Incubation period 86 days, 214 t.u. Average temperature 1.7° C.*

FIG. 25. *Surface view of developing egg. Length of embryo 13.5 mm. Series I. St. John Hatchery, N.B., Mar. 15, 1935. Incubation period 128 days, 256 t.u. Average temperature 1.2° C.*

FIG. 26. *Surface view of developing egg. Length of embryo 15.8 mm. Series I. St. John Hatchery, N.B., Apr. 12, 1935. Incubation period 156 days, 284 t.u. Average temperature 1.1° C.*

FIG. 27. *Surface view of developing egg. Length of embryo 18.6 mm. Series I. St. John Hatchery, N.B., Apr. 26, 1935. Incubation period 170 days, 310 t.u. Average temperature 1.1° C.*

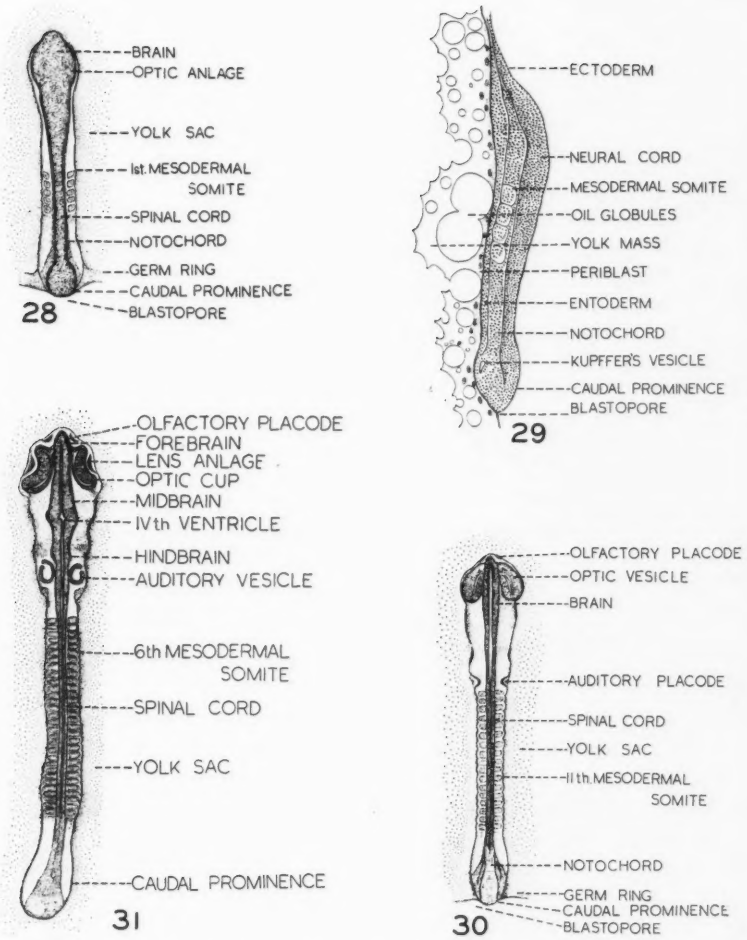


FIG. 28. *Dorsal view of embryo, enlarged. Length 2.1 mm., five somites. Series I. St. John Hatchery, N.B., Nov. 29, 1934. Incubation period 22 days, 129 t.u. Average temperature 3.4° C.*

FIG. 29. *Slightly oblique sagittal section of embryo. Length 2.0 mm., four somites. Series I. St. John Hatchery, N.B., Nov. 29, 1934. Incubation period 22 days, 129 t.u. Average temperature 3.4° C.*

FIG. 30. *Dorsal view of an embryo, enlarged. Length of embryonic axis 3.3 mm., 17 somites. Series III. Birch Cove Hatchery, N.B., Jan. 24, 1940. Incubation period 68 days, 109 t.u. Average temperature 1.5° C.*

FIG. 31. *Dorsal view of embryo. Total length 4.2 mm., 30 somites. Series III. Birch Cove Hatchery, N.B., Feb. 5, 1940. Incubation period 80 days, 120 t.u. Average temperature 1.3° C.*

FIG. 32. *Lateral view of embryo. Total length 4.8 mm., 40 somites. Series I. St. John Hatchery, N.B., Dec. 13, 1940. Incubation period 36 days, 164 t.u. Average temperature 2.5° C.*

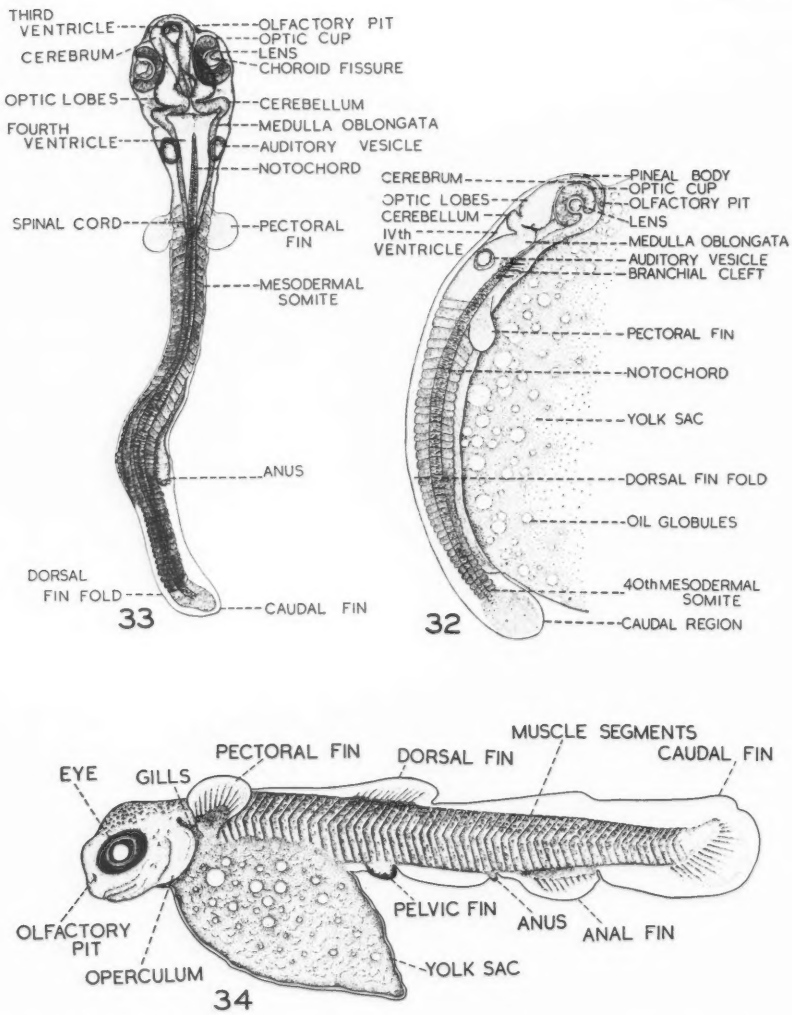
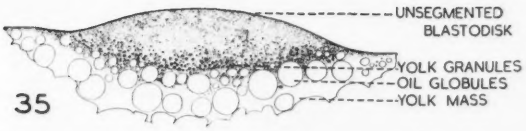
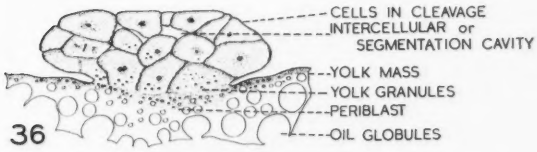
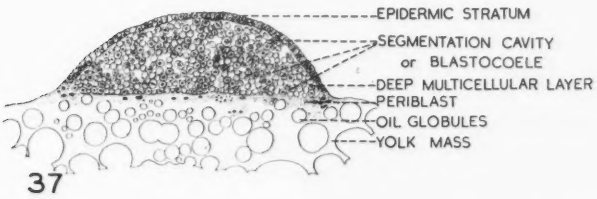


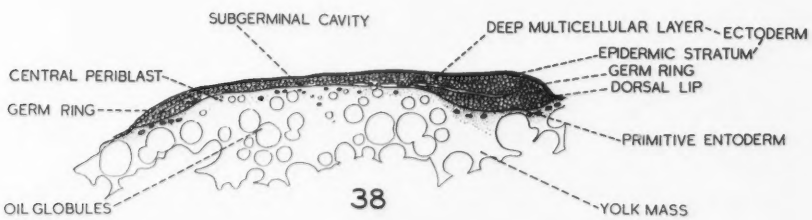
FIG. 33. *Dorsal view of embryo. Length 6.0 mm., 60 somites. Series III. Birch Cove Hatchery, N.B., Feb. 20, 1940. Incubation period 95 days, 136 t.u. Average temperature 1° C.*

FIG. 34. *Newly hatched larva. Total length 22 mm. Series I. St. John Hatchery, N.B., May 10, 1935. Incubation period 181 ± 3 days, 470 t.u. Average temperature 1.6° C.*









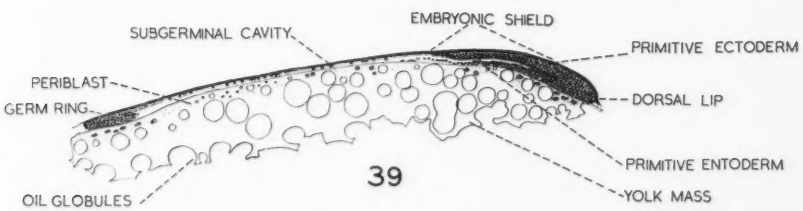


FIG. 35. *Transverse section of blastodisk of recently fertilized egg. Diameter of blastodisk 1.28 mm. Series II. A. B. S., Nov. 2, 1934. Seven hours after fertilization. Temperature 7° C.*

FIG. 36. *Transverse section of blastodisk of diameter 1.4 mm. Series II. A. B. S., Nov. 4, 1934. Forty-one hours after fertilization. Temperature 7.5° C.*

FIG. 37. *Transverse section through late segmentation stage. Diameter of blastodisk 1.48 mm. Series II. A. B. S., Nov. 6, 1934. Incubation period four days, 59 t.u. Average temperature 8.4° C.*

FIG. 38. *Sagittal section of blastoderm showing formation of early subgerminal cavity. Diameter through embryonic shield 2.6 mm. Series II. A. B. S., Nov. 10, 1934. Incubation period eight days, 127 t.u. Average temperature 8.4° C.*

FIG. 39. *Sagittal section through blastoderm showing subgerminal cavity becoming gradually obliterated. Diameter of blastoderm 3.85 mm. Series II. A. B. S., Nov. 12, 1934. Incubation period 10 days, 138 t.u. Average temperature 8° C.*

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A SYSTEMATIC STUDY OF THE MAIN ARTERIES IN THE REGION OF THE HEART—AVES XXIII

PASSERIFORMES (COMPSOTHLYPIDAE)[1]

By Fred H. Glenny[2]

Abstract

Ten species of the Compsothlypidae were dissected and diagrams of the arterial arrangement in the neck and thorax prepared. The basic family arrangement-pattern was found to be characteristic for all of the species studied. The ligamentum aortae was present and somewhat reduced, while the right ligamentum botalli was completely atrophied. The intercostal arteries arose near the base of the coracoid major arteries.

Introduction

The Compsothlypidae, like other families of the Passeriformes (1, 2, 9, 10, 11) are *aves bicarotidinae normales.*

While many families of the Passeriformes have basic arterial arrangement-patterns that are compact or well-defined, still others do not present definite well-defined characteristic family arrangement-patterns. As the writer has pointed out on previous occasions, well-defined ordinal or family patterns (by which orders, families, and subfamily groups may be further divided or differentiated) appear to indicate trends of divergence in the evolution of the family or order. Families and orders that present a high degree of uniformity in the arrangement of the arteries in the neck and thorax probably represent groups in which wide divergence may no longer be expected to take place—and as a result these same groups may well represent terminal groups or end-lines in this limited phase of their evolution.

While it is seldom found that whole orders of birds—other than those that are represented by but one family and but a very limited number of species—present arterial patterns that indicate terminal evolution, it is not uncommon to find families within these orders that do appear to be terminal forms in the evolution of the order. This is particularly noticeable in the various families of the Passeriformes.

Materials

The materials that were included in this study were collected by the Cleveland Museum of Natural History and by the author and consisted of the following species of birds. Unless otherwise indicated, only single specimens were dissected.

Dendroica cerulea Linnaeus
Dendroica coronata Linnaeus

[1] *Manuscript received May 10, 1944.*
Contribution from the Department of Zoology, University of Toronto, Toronto, Ont., Canada.

[2] *Formerly Assistant, Department of Zoology, University of Toronto. Now on Active Service with the United States Army.*

Dendroica fusca Müller
Geothlypis trichas trichas Linnaeus
Oporornis formosus Wilson
Protonotaria citrea Boddaert
Seiurus aurocapillus Linnaeus
Setophaga ruticilla Linnaeus (two specimens)
Vermivora rubricapilla Wilson
Vermivora peregrina Wilson (two specimens)

Observations

The arrangement-pattern of arteries in the neck and thorax of the Compsothlypidae (Fig. 1) is characteristic for the species examined in this study. Only very minor individual (non-specific) variations were observed in any of the specimens.

The innominate artery (*2*) divides to give rise to the common carotid (*7*) and subclavian (*8*) arteries. The subclavian then gives rise to the coracoid major (*9*), axillary (*11*), and two pectoral (*12*) arteries in order. The intercostal (*10*) arises as a branch of the coracoid major artery.

The common carotid gives rise to the thyroid artery (*13*) and ductus shawi (*14*) before dividing to form the cervicovertebral (*21*) and internal carotid (*19* and *20*) arteries. The ductus shawi (*14*) sends off a syringotracheal artery (*15*) before passing posteriorly. The cervicovertebral artery (*21*) divides to form the vertebral (*16*) and superficial cervical (*17*) arteries. The scapular artery (*18*) arises as a branch of the superficial cervicals. The left internal carotid (trunk) artery (*19*) alone enters the hypapophysial canal, while the right complementary vessel has become functionally modified to serve as the ascending-oesophageal artery (*20*) of the adult.

The ligamentum aortae (*5*) maintains its proximal attachment with the pulmonary artery (*6*) and its distal attachment with the right radix aortae (*4*) just anterior to the dorsal aorta. The right systemic (fourth aortic) arch (*3*) arises from the right innominate artery near its origin from the aortic root (*1*).

Discussion

From the present studies, the writer concludes that the Compsothlypidae represent a well-defined group of birds that may be considered to present major terminal evolution characteristics. This is particularly noticeable with regard to the evolution—especially with regard to divergences—in the aortic arches and their derivatives. One of these chief characteristics may be based upon the extremely similar arrangement-patterns of the arteries of the neck and thorax of the different species of this family. In as much as these vessels are chiefly derivatives of the embryonic aortic arches and certain other embryonic vessels, it may be assumed that were there any predisposition toward divergence within the family—as appears to be the case within the *Corvidae*—such trends in evolutionary divergence might well be observed in

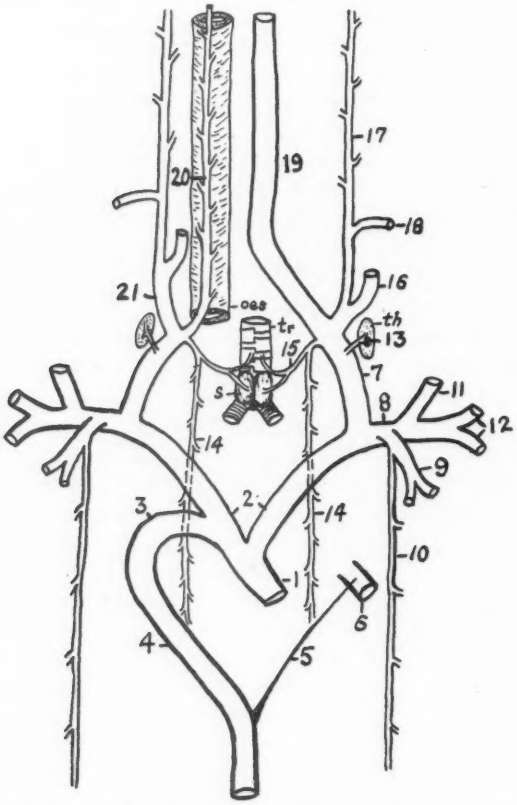

FIG. 1. *Main arteries of the neck and thorax of Seiurus aurocapillus. Ventral view.*

KEY TO ABBREVIATIONS

(*1*) Aortic root; (*2*) innominate arteries; (*3*) right aortic arch; (*4*) right radix aortae; (*5*) ligamentum aortae; (*6*) pulmonary artery; (*7*) common carotid artery; (*8*) subclavian artery; (*9*) coracoid major artery; (*10*) intercostal artery; (*11*) axillary artery; (*12*) pectoral arteries; (*13*) thyroid artery; (*14*) ductus shawi; (*15*) syringotracheal artery; (*16*) vertebral artery; (*17*) superficial cervical artery; (*18*) scapular artery; (*19*) internal carotid (trunk) artery; (*20*) ascending-oesophageal artery; (*21*) cervicovertebral artery; oes. = oesophagus; tr. = trachea; s. = syrinx; th. = thyroid gland.

the arterial arrangement-patterns, which in turn appear to have some phylogenetic significance.

The phylogenetic significance of the pattern of the arteries in the neck and thorax is more readily observable in the different orders of birds—and particularly in the Piciformes (*4*), Coraciiformes (*3*), Anseriformes (*5*), Psittaciformes (*1*), Galliformes (*7*), Tinamiformes (*6*), and Ciconiiformes (*8*)—than is evident, at the present time, in other classes of Vertebrata. It is entirely possible that other classes of vertebrates may present characteristic

ordinal and perhaps even family arrangement-patterns of the arteries derived from the embryonic aortic arches and vertebral arteries. There appears to be no organized systematic study of this kind on any other class or order of vertebrates, however, so that further conclusions as to the importance of this feature cannot be drawn with regard to other classes. It remains a characteristic of some importance, however, in the study of lines of evolution among the birds.

Acknowledgments

The writer wishes to express his gratitude to Dr. E. Horne Craigie, F.R.S.C., Department of Zoology, University of Toronto, Dr. Harry C. Oberholser, Cleveland Museum of Natural History, and Dr. Alexander Wetmore, Assistant Secretary, Smithsonian Institute for assistance in obtaining specimens for this study and for the many helpful suggestions and comments in the preparation of this paper.

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